Parasitism of the eggs of *Lygus shulli* and *Lygus elisus* (Heteroptera: Miridae) by *Anaphes iole* (Hymenoptera: Mymaridae)

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ABSTRACT

Females of the egg parasitoid *Anaphes iole* Girault (Hymenoptera: Myrmaridae) accepted and oviposited in eggs of both *Lygus shulli* Knight and *L. elisus* Van Duzee (Heteroptera: Miridae) when presented on sections of green bean pod in the laboratory. Resulting *A. iole* larvae developed normally on eggs of both host species and emerged as adults. The wings of *A. iole* emerging from *L. shulli* eggs were significantly larger than those from *L. elisus* probably because the eggs of *L. shulli* were larger. *Anaphes iole* females parasitized only approximately 50% of the eggs available of either host species. This may indicate that 50% of the hosts were suitable and rejected, that 50% were unsuitable for oviposition, or that the structure of bean pods prevents females from finding or ovipositing in 50% of hosts. *Anaphes iole* has potential for biological control of *Lygus* spp. on greenhouse vegetables in southwestern British Columbia.

Key words: *Anaphes iole*, Mymaridae, *Lygus shulli*, *Lygus elisus*, *Lygus hesperus*, Miridae, egg parasitoids, biological control, greenhouse vegetables

INTRODUCTION

Plant bugs in the genus *Lygus* (Heteroptera: Miridae) feed on a wide variety of native plant species and agricultural crops throughout North America (Schwartz and Footit 1992; Schwartz and Footit 1998). Sequential migration among different seasonally-occurring host-plant species is typical of the biology of *Lygus* (Schwartz and Footit 1992). As a consequence, *Lygus* species migrate into, and feed upon, acceptable agricultural crops. Economic damage occurs when *Lygus* nymphs or adults feed on reproductive tissues or apical meristems (Schwartz and Footit 1992). Damage has been reported on cotton (Leigh *et al.* 1988), alfalfa (Sorenson 1936), canola (Butts and Lamb 1991; Wise and Lamb 1998), strawberries (Norton and Welter 1996) and conifer seedlings (Schowalter 1987). In southwestern British Columbia (BC), *Lygus* bugs are sporadically-occurring pests of greenhouse vegetable crops like cucumber and sweet pepper (Gillespie *et al.* 1999). Here, we report on parasitism of the eggs of *L. shulli* Knight and *L. elisus* Van Duzee by the egg parasitoid *Anaphes iole* Girault (Hymenoptera: Mymaridae), and discuss the potential of *A. iole* for biological control of *Lygus* species in BC vegetable greenhouses.

Lygus hesperus Knight, *L. shulli* and *L. elisus* are the three most common species of *Lygus* in the Fraser Valley of southwestern BC (Gillespie *et al.* 1999). *Lygus shulli* and *L. elisus* are sporadically found in vegetable greenhouses throughout the growing season, and their feeding damages both cucumbers and peppers (Gillespie *et al.* 1999). *Lygus hesperus* occurs only on late-season pepper crops, and there is no definitive evidence, at present, that

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this species causes economic damage (Gillespie *et al.* 1999). Invasion of greenhouses probably occurs when *Lygus* bugs disperse from surrounding weedy habitat (Gillespie *et al.* 1999).

Augmentative releases of the egg parasitoid *Anaphes iole* have been used to manage populations of *L. hesperus* in field plantings of strawberries in California (Norton *et al.* 1992; Norton and Welter 1996; Udayagiri and Welter 2000). Although *A. iole* is available commercially (Biotactics Inc., Riverside, California) and could potentially be used for biological control of *Lygus* spp. in BC, no information is currently available regarding parasitism of *L. shulli* and *L. elisus* by this parasitoid. In this paper, we report on the acceptance and suitability of *L. shulli* and *L. elisus* eggs as hosts for *A. iole*.

MATERIALS AND METHODS

Host eggs for parasitism bioassays. Adults of *L. shulli* and *L. elisus* were collected at field sites near the Pacific Agri-Food Research Centre in Agassiz, BC in July 1999. *Lygus* adults were identified to species using Henry and Froeschner (1988). Adults of each species were held at ambient conditions in the laboratory in groups of 5-10 in cylindrical 500-ml plastic containers with screened lids. *Lygus* adults were provided with 5-10 sections of green bean (3 to 5 cm in length) for feeding and oviposition. Bean sections were inspected daily for *Lygus* eggs and those with two or more eggs present were removed from the containers and stored in a refrigerator at 10°C until used in parasitism bioassays. Bean sections were replaced in oviposition containers when removed (as above) or when their condition deteriorated (*e.g.* showing signs of fungal growth).

Parasitism bioassays. Adult Anaphes iole of unknown age were obtained directly from a commercial supplier (Biotactics Inc., Riverside, California). Female *A. iole* were selected for use in oviposition bioassays by examining abdominal morphology under a dissecting microscope to determine sex. Individual female *A. iole* were introduced using a moistened fine paintbrush into 62-ml plastic cups (Solo Cup No. P200, Solo Cup Company, Urbana, Illinois) each containing a single section of green bean with eggs of either *L. shulli* (91 cups) or *L. elisus* (82 cups). The number of eggs available for parasitism on each bean section ranged from 2 to 36 for *L. elisus* and from 2 to 68 for *L. shulli*. The cups were held in a growth chamber for 24 h to allow *A. iole* to oviposit (L:D 16:8 h photoperiod and temperatures of 23.0 ± 0.2°C (±SD) during photophase and 18.0 ± 0.2°C (±SD) during scotophase; relative humidity was not controlled). After 24 h, the female *A. iole* were removed with a moistened paintbrush and the cups were returned to the growth chamber and checked every second day for emergence of *Lygus* nymphs or *A. iole* adults until no further emergence occurred. Emerging *Lygus* nymphs were removed, counted and discarded. Emerging *A. iole* adults in each cup were removed and stored separately, as families, in 70% (w/v) ethanol. The total numbers of *Lygus* nymphs and *A. iole* males and females that emerged from each bean section were recorded.

Wing size of *A. iole* adults. The right wing of *A. iole* adults that emerged from bean sections was measured as an index of body size. From the offspring stored in ethanol, we selected at random, one male and one female adult from each of 32 randomly-selected families reared on *L. shulli* and 31-32 families reared on *L. elisus*. The length (from the point of attachment to the tip of the wing) and width (at the widest point) of each right forewing were measured and recorded.

Size of *Lygus* eggs. Adults of *L. shulli* and *L. elisus* were collected at field sites near the Pacific Agri-Food Research Centre at Agassiz, BC in July of 2000. The ovaries of 10 adult females of each species were dissected. The length (from tip to tip) and width (at the widest point) of all mature, fully chorionated eggs for each female were measured and recorded.

Data analysis. Total number of viable host eggs in parasitism bioassays was calculated as the totals of *Lygus* and *A. iole* emerging from each bean section. This calculation gives a conservative estimate of the original number of *Lygus* eggs per bean section as it assumes that no *Lygus* or *Anaphes* eggs died during the test. The proportion of eggs parasitized per female was calculated as the number of *A. iole* emerging divided by the total number of host eggs. The proportion of female *A. iole* emerging from each bean section (sex ratio) was calculated as the number of females emerging divided by the total of both sexes (F/(F+M)). Proportion of *Lygus* eggs parasitized, number of *A. iole* emergents, number of *Lygus* emergents, total number of host eggs and sex ratio of *A. iole* emergents (all per bean section) were compared between *L. shulli* and *L. elisus* using Mann-Whitney tests. Proportional data were arcsin-square root transformed before analysis. The length and width of the wings of male and female wasps emerging from *L. shulli* and *L. elisus* were compared using Mann-Whitney tests. The length and width of the eggs of *L. shulli* and *L. elisus* were compared using Mann-Whitney tests. All statistical analyses were conducted using Systat (Wilkinson *et al.* 1997) and Sigmastat (Fox *et al.* 1995).

RESULTS

Parasitism bioassays. Female *A. iole* oviposited in eggs of both *Lygus* species, and both species were suitable for the development of *A. iole* from larva to adult. There was no significant difference in the proportion of eggs parasitized per *A. iole* female for either *L. elisus* or *L. shulli* (Table 1). The number of *A. iole* and *Lygus* emerging and the total number of host eggs per bean section were significantly higher for *L. shulli* than for *L. elisus* (Table 1). Previous observations indicate that *L. shulli* is more likely to oviposit on green beans than *L. elisus* (D.M.J. Quiring, personal observation). There was no significant difference in the sex ratio of *A. iole* emerging from *L. shulli* compared to *L. elisus* eggs (Table 1).

Table 1

Proportion of *Lygus* eggs parasitized, number of *Anaphes* and *Lygus* emerging, number of *Lygus* host eggs, and sex ratio (Means ± SE) of *A. iole* emerging per bean section containing host eggs of *L. shulli* and *L. elisus*.

Variable	*L. shulli*	*L. elisus*	Mann-Whitney test U	*P*
Proportion of eggs parasitized	0.46 ± 0.04 (n=91)	0.49 ± 0.04 (n=82)	3952	0.50
Number of *Anaphes* emerging	10.4 ± 1.1 (n=91)	5.1 ± 0.5 (n=82)	2986	0.01
Number of *Lygus* emerging	10.9 ± 1.0 (n=91)	6.0 ± 0.6 (n=82)	2549	< 0.001
Total number of host eggs	21.2 ± 1.3 (n=91)	11.1 ± 0.8 (n=82)	1791	< 0.001
Sex ratio (F/(F+M))	0.55 ± 0.03 (n=64)	0.52 ± 0.04 (n=67)	1991	0.48

Wing size of *A. iole* adults. The mean length and width of the right forewings of both male and female *A. iole* emerging from the eggs of *L. shulli* were significantly greater than those emerging from the eggs of *L. elisus* (Table 2).

Table 2

Length and width (Mean ± SE) of right forewings of *Anaphes iole* males and females reared on the eggs of *Lygus shulli* and *Lygus elisus*.

Variable	*L. shulli*	*L. elisus*	Mann-Whitney test U	*P*
Female wing length (mm)	0.75 ± 0.01 (n=32)	0.71 ± 0.01 (n=32)	204	< 0.001
Female wing width (mm)	0.17 ± < 0.005 (n=32)	0.15 ± < 0.005 (n=32)	202	< 0.001
Male wing length (mm)	0.79 ± < 0.005 (n=32)	0.75 ± 0.01 (n=31)	133	< 0.001
Male wing width (mm)	0.18 ± < 0.005 (n=32)	0.17 ± < 0.005 (n=31)	343	0.033

Size of *Lygus* eggs. The mean length of *L. shulli* eggs (0.99 ± 0.01 mm) was significantly greater than that of *L. elisus* eggs (0.91 ± 0.01 mm; Mann Whitney test: U = 132, *P*<0.001), as was the mean width of *L. shulli* eggs (0.26 ± 0.00 mm) compared to *L. elisus* eggs (0.23 ± 0.00 mm; Mann Whitney test: U = 224, *P*<0.001).

DISCUSSSION

Anaphes iole females readily accepted, and oviposited in, eggs of both *L. shulli* and *L. elisus*. Their offspring developed successfully in both species of host eggs and emerged as adults. *Anaphes iole* are available commercially and readily parasitize all three *Lygus* spp. (*L. shulli*, *L. elisus*, and *L. hesperus*) found in greenhouses in the Fraser Valley of southwestern BC. *Anaphes iole* thus has a strong potential for biological control of *Lygus* spp. in BC vegetable greenhouses.

No significant differences were found between the two host species in the proportion of available eggs parasitized by *A. iole* females. The proportion of eggs parasitized was approximately 50% for both species despite the fact that, on average, nearly twice the number of host eggs were oviposited on bean sections by *L. shulli* compared with *L. elisus*. *Anaphes iole* females typically carry between 30 and 40 mature eggs in their abdomens, and an individual female can parasitize approximately 30 *Lygus* eggs per day (S. Udayagiri, University of California, personal communication). Assuming that no *A. iole* eggs died after oviposition in our experiments, the *A. iole* females in this study oviposited substantially fewer eggs (on either host species) during 24 h than they had available (5 eggs on average on bean sections with *L. elisus* eggs and 10 eggs on average on those with *L. shulli* eggs). It is possible that 50% of the eggs of both species were either suitable hosts that were rejected by females, or unacceptable hosts for oviposition. Alternatively, 50% of *Lygus* eggs could have been inaccessible for oviposition, or impossible to locate, by *A. iole* on bean sections. Recently, it has been shown that plant structure can influence the oviposition success of *A. iole*. A lower proportion of *Lygus* eggs were parasitized by *A. iole* females on strawberry fruits than on petioles, leaflets or calyx tissue (Udayagiri and Welter 2000). Achenes (one-seeded fruitlets) on strawberry fruits apparently hinder access by *A. iole* females to *Lygus* eggs present on fruits (Udayagiri and Welter 2000). If *A. iole* is to be used for biological control of *Lygus* spp. in BC greenhouses, it will be critical to determine whether plant structure affects the ability of *A. iole* females to locate and parasitize eggs on cucumbers and peppers.

Anaphes iole adults that emerged from *L. shulli* eggs had larger wings than adults from *L. elisus* eggs. Assuming that wing size correlates with body size, *L. shulli* eggs may be of higher quality for development of *A. iole* than *L. elisus* eggs. This is likely a consequence of the larger size of *L. shulli* eggs compared to *L. elisus* eggs. Alternatively, the difference in wing size we observed may be caused by some influence of the host not related to egg size. Host-induced variation in antennal morphology unrelated to host size was found in *A. iole* by Huber and Rajakulendran (1988). Determining whether wing-size variation between hosts reflects differences in host-egg size or other effects will require further research.

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